

Observations on the zoospores of *Draparnaldia*.

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WITH PLATE XXXII.

Having been interested during the past season in studying various algæ I have been surprised at the lack of accurate descriptions of the zoospores and their mode of formation. Aside from the familiar papers on *Ulothrix zonata* little seems available, beyond scattered references in various text books.

The plant selected for special study was *Draparnaldia plumosa* Ag. Concerning this genus little information can be gleaned from the accessible books. The accounts by the older writers are chiefly interesting historically. For instance Decaisne¹ in 1842 spoke briefly of the zoospores, and figures them though with no attempt at detail. He says that he failed to see any motion of them either before or after their escape from the cell. It is possible that he saw only resting spores.

Kuetzing, in his "Metamorphose des vegetaux inferieurs,"² mentions finding *Draparnaldia* producing zoospores on one occasion, but lost the material before making any detailed study of them. He speaks with regret of thus failing to have the opportunity to investigate the vexed question whether zoospores were really animals. Before this Treviranus had observed the zoospores and shown that from them grew young plants of *Draparnaldia*.

Derbes and Solier, in a paper published in 1850,³ gave a brief but concise account of the zoospores in the closely related *Stigeoclonium tenue* Kg.

Aside from these references there is nothing available, at least to the average botanist. This being so it has been thought desirable to give a somewhat minute account of the structure and formation of the zoospores in the species mentioned.

The form of the plant is probably familiar to every botanist. The main axis is comparatively stout, and is made up of cylindrical or barrel shaped cells, with the walls lined by a

¹Essai sur une classification des algues, etc. Annales des Sc. Nat. Bot. II. xvii. 314.

²Annales des Sc. Nat. Bot. II. xi. 136.

³Organes reproducteurs des algues. Ann. des Sc. Nat. Bot. III. xiv. 267.

central transverse band of chlorophyll, containing several pyrenoids. On the sides of this axis and its main branches are borne numerous smaller branchlets, each itself branching copiously into a plumose tuft. The cells of these branchlets are shorter and their chlorophyll covers nearly the entire wall. Toward the end of the ultimate branchlets the cells are much longer, with the narrow chlorophyll band central. The young branchlet is tipped with a long hyaline multicellular seta.

The production of zoospores is confined to the branchlets, but to no particular part of them. Often the contents of all the cells of a branchlet become thus transformed, and it is not uncommon to find a vigorous plant changed in a single day to a naked axis surrounded by the empty cell walls of its branchlets.

The zoospore is ovate or oval in form (fig. 1) and averages about $12-16 \times 8-10 \mu$, though there is much variation in both form and size. The anterior end is rather pointed and nearly hyaline. On the tip are attached the four cilia, each rather longer than the body. The chlorophyll is parietal, but not uniformly distributed, and contains one or more pyrenoids. There is a bright red pigment spot, lanceolate in outline and apparently slightly raised above the surface. It is usually placed about on the middle of the side of the zoospore, in a diagonal position. It is always connected with the chromatophore, and in one case was observed at the very base of the body. At the anterior end of the zoospore there are two large contractile vacuoles, placed side by side and contracting alternately. Repeated observations showed that the average interval between two contractions of the same vacuole was about fifteen seconds. After contraction the vacuole often seemed invisible for about six seconds, then appeared and expanded gradually for ten to twelve seconds, when it suddenly and rapidly contracted. Occasionally the contraction was slower, occupying nearly a second.

Only a single zoospore is formed in each cell. The first sign of their production is apparently the formation of the pigment spot. This seems to be fully formed at least twenty-four hours before the escape of the mature zoospore, but the manner of its formation could not be determined. One can often find a branchlet differing apparently in no respect from those around it, except that each cell bears in some part of

its interior this bright red spot. This is especially striking when seen in one of the long terminal cells where the chromatophore lines but a small portion of the cell wall. In some cases the cells at this stage seem to have the contents slightly more granular than usual. The contents continue ordinarily to occupy the whole cavity of the cell wall up to the very instant of escape, though occasionally the zoospore assumes a somewhat rounded form while still within the mother cell. It is difficult to determine the exact time of formation of the vacuoles, but in every case where a vacuole was observed pulsating the zoospore escaped in less than fifteen minutes.

The escape of the zoospores takes place normally in the morning, and most abundantly from eight to ten o'clock, though not infrequently occurring at any hour of the day. Specimens collected in cold weather and brought into a warm room soon begin to discharge their zoospores in abundance, regardless of the time of day. They seem to be formed at all times of the year, having been observed by the writer in nearly every month.

There is usually no motion or change in the appearance of the zoospore to denote that the time of escape is at hand. Suddenly the wall gives way at one point, and through the opening bursts the zoospore, the hyaline end being invariably first to appear (fig. 2). The whole zoospore may escape almost instantly, or the process may be prolonged for at least a quarter of an hour. In a few cases the zoospore was seen with its anterior end protruding slightly from the small opening, and vigorously twisting, with a boring motion, apparently trying to force its way out. If the opening is not large enough for easy escape, the zoospore assumes a dumb-bell shape, the body gradually squeezing through. Figures 3 and 4 represent a specimen in which the escape was rather rapid. Just as the first drawing was finished there was a sudden bursting of all the cells, and every zoospore thrust its colorless end out. The other drawing (4), showing the same cells two minutes later, was hastily outlined, and in four minutes every cell was empty.

The zoospore is often held even after its body is free, by its cilia, which seem to get caught either inside the cell or by the mucilage covering it. Then follow violent struggles to get free. The opening in the cell wall seems to be produced, in part at least, by absorption of its substance, for no projecting edges can be seen.

As soon as the zoospore is free it darts rapidly away, with a rotary motion, and keeping the hyaline end forward. Under the microscope it never moves far in the same direction, but seems to turn at random. It is almost impossible to determine with any degree of accuracy the rate of their motion, but the result of repeated trials indicates an average velocity of about 6^{mm} per minute.

The period of activity does not seem to be very long—in no case observed longer than seven minutes. Judging from the distance to which they sometimes travel it must often be rather longer than this. At the end of the period the motion diminishes in velocity, and the body begins to rotate around the hyaline end. This motion soon grows slower, and after a few final quivers it ceases. In settling down the zoospores show a tendency to collect in groups like that shown in figure 5, or to attach themselves by their hyaline ends to bits of gravel, the larger filaments of the plant, or in fact any object which comes in their way. One often sees old plants completely covered by the masses of zoospores fastened to them. No signs of conjugation have been seen, but most of the zoospores germinate freely.

The contraction of the vacuoles ceases soon after the zoospore ceases its motion, and the cilia disappear. The cell quickly begins to elongate and a slight disc is formed on the hyaline end, by which it clings to the substratum. A cell wall forms around the hitherto naked mass, and the free end of the cell becomes pointed as it elongates. The elongation takes place above the pigment spot, leaving this near the point of attachment. The first cell division may take place in a few hours or may be delayed for several days. This divides the spore in such a way that the pigment spot always remains in the basal cell (fig. 7). It may persist till four or five cells have been formed (fig. 9), when it begins to grow fainter, and seems to be absorbed. About this time the basal cell begins to form an hyaline filament, which grows rapidly over the substratum, and serves as a hold-fast for the young plant.

By the time the filament has reached a length of ten or twelve cells the tip has elongated into a multicellular bristle, which growing rapidly soon becomes several times as long as the filament (figs. 10, 11). Of course the growth of the filament must then be intercalary. From this point the young

plant grows rapidly, and soon becomes large enough to begin the formation of another generation of zoospores, which it does almost before the main axis has been developed.

One of the most interesting and striking peculiarities of the zoospores is their behavior toward light. It has been said that they appear chiefly in the morning, but this is apparently due, in part at least, to the change of temperature, for it was proved by experiment that they were produced quite as abundantly in a perfectly dark box as in the bright light outside. The effect of the darkness seemed to be to slightly retard the time of formation, making its maximum about ten o'clock in the morning (in winter). The optimum temperature of the water seems to be about 17° C.

Like most other zoospores these are heliotropic, swarming toward the light in such numbers that the side of the vessel becomes coated with them. Various experiments were performed to determine whether this action was really due to the light or to the heat. The result of these experiments was to show that it is the light rays which influence them, though in some cases it seemed that the zoospores were slightly affected by the heat.

It may be well in closing to state that in autumn many plants were found whose filaments were transformed into chains of resting spores. These differed little in general appearance from zoospores, excepting that they lacked the pigment spot and vacuoles, and that their contents seemed uniformly distributed in a parietal layer. This was nearly homogeneous and contained several pyrenoids. They do not seem to leave the parent cells, but germinate, the shoot bursting through their wall. The appearance of these resting spores suggests that they are modified zoospores, but in none could any pigment spot be detected or any other proof of such an origin.

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EXPLANATION OF PLATE XXXII.

Figures 1-8 \times 1150. Figures 9-11 \times 850. All reduced one-third in engraving.

Fig. 1. Mature zoospores.—Fig. 2. Filament, showing zoospores in the act of escaping.—Figs. 3, 4. Two cells showing condition before and during escape of zoospores. *v*, vacuole. *p*, pigment spot.—Fig. 5. Cluster of zoospores showing characteristic grouping as their motion ceases.—Figs. 6-11. Showing growth of young plant from zoospore.